Abstract

A method for the identification of the diarrheagenic E. coli groups: ETEC (enterotoxigenic E. coli), A/EEC (attaching and effacing E. coli) EPEC (enteropathogenic E. coli), VTEC (verocytotoxin producing E. coli) and EIEC (enteroinvasive E. coli), and Shigella spp. is described. The bacterial identification is made possible by the specific detection of the following virulence genes: sta and elt encoding heat stable enterotoxin (ST) and heat labile enterotoxin (LT) characteristic of ETEC, eae encoding intimin, characteristic of A/EEC, EPEC or VTEC, bfpA encoding bundle forming pilus (BfpA), characteristic of EPEC, vtx1 and vtx2 encoding veroxytotoxin 1 and 2 (VT1 and 2) characteristic of VTEC, ipaH encoding invasive plasmid antigen H (IpaH) characteristic of EIEC and Shigella spp., and ehxA encoding enterohemolysin (EhxA) characteristic of some EPEC and VTEC strains. The method allows the simultaneous detection of any combination of the 8 virulence genes by one single multiplex-PCR. The method is thoroughly validated with respect to sensitivity and specificity, and showed high performance compared to other publication. The method includes an internal positive PCR control and the carry-over prevention system, UNG, which makes it ideal for routine diagnostic analyses. The method can be combined with a number of other technologies leading to even higher sensitivity and reduced time of analysis - both important parameters when diarrheagenic patient or contaminated foods are analyzed.